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## PRODUCTION OF IMMUNE SERUM FOR STREPTOCOCCUS ALPHA PRIME\*

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A case of subacute bacterial endocarditis, proved by blood culture, was admitted to the Lankenau Hospital on the service of Dr. Fred L. Hartmann, July 3, 1938. The patient had been admitted to another hospital in April, 1938. At that time the blood culture was found to be positive. Streptococcus was the offender. Its peculiar type was not described. The patient received sulfanilamide with no decrease in bacterial count and she developed a sensitivity to the drug. After several weeks' stay, the patient left the hospital and was admitted to the Lankenau Hospital.

The first blood culture was taken July 4, 1938. A 1% dextrose infusion broth was used. About 8 cc. of the blood was added to 50 cc. of this broth. One cc. of the blood was added to each of two tubes of agar-agar, melted and cooled to 42 C. The contents of these two tubes were well mixed and were then poured into sterile

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Petri plates. After three days' incubation, a uniform chain formation of streptococci was found in a smear made from the flask. The plates showed four colonies per 1 cc. of blood on the same day. The colonies were small with an area of partial hemolysis about one and a half times the diameter of the colony itself. The area of hemolysis was not clear cut as in the true *Streptococcus hemolyticus* nor did it present the green zone of methemoglobin, typical of *Streptococcus viridans*. Since this was my first encounter with such an organism, I consulted Dr. Harry E. Morton, a member of the bacteriological department of the U. of Penna. Medical School. He expressed his opinion that the organism belonged to the alpha prime group and suggested that I present my problem to Mr. Roos, bacteriologist at Sharp and Dohme.

Mr. Roos requested a culture of the organism which he tested with approximately fifty strains of streptococci. These strains represented the hemolytic, viridans and some of the intermediate types with no evidence of agglutination in any of them. Since our aim was to give immune serum, if possible, Mr. Roos suggested that I immunize rabbits with the original organism. Since this work was entirely new to me, he gave me directions for planning the work which I followed very carefully.

The culture was adapted to infusion broth by frequent subcultures until the growth was quite heavy after six hours' incubation. This procedure took about two days. One per cent formalin was added to a fresh 6 hr. culture and it was allowed to stand at room temperature for 24 hrs. After centrifugation at high speed for 15 mins., the supernatant fluid was decanted and the bacterial sediment was resuspended in Locke's solution containing 0.1% formalin. From this heavy suspension, the necessary dilutions were made as they were needed. The strength of the organisms was determined with an nephelometer. The sterility of the suspension was tested. On July 22, 1938, the first rabbit was injected. One cc. of Locke's solution containing 500,000,000 of bacteria was injected into the rabbit, intravenously. Twice a week the rabbit received an increasing dose of the bacterial suspension. The strength increased as follows: 1,000,000,000 — 2,000,000,000 — 3,000,000,000 — 4,000,000,000 — 5,000,000,000, respectively. Four days after the last injection, the rabbit was bled from the marginal ear vein. A healthy uninoculated rabbit was bled at the same time as a control.

Using a very heavy suspension of bacteria in saline, the Huddleson type of macroscopic agglutination was used. These organisms were not treated with formalin because of the possibility of false agglutination. The suspension was heated at 56 C. for 2 hrs. for the test. The bacterial suspension represented approximately, 20,000,000,000 organisms per 1 cc. A series of dilutions of serum from the inoculated rabbit and the control rabbit were made in a porcelain test plate, ranging from 1:10 to 1:5,120. A piece of glass plate 4 x 7 inches was divided into  $\frac{3}{4}$  inch squares with a red china marking pencil. One drop of the above mentioned dilution of serum was placed in each square. To it was added one drop of the heavy suspension of organisms. The Wright pipettes used for these drops were approximately the same calibre. Each mixture was thoroughly agitated with an applicator. The entire plate was rocked and rotated for approximately 2 mins., read and shaken again for one more minute, using the final reading obtained, after the second agitation. By tilting the plate slightly before a microscope lamp, placed on a dark background, the agglutination could be seen very clearly, macroscopically. The weaker dilutions showing doubtful agglutination were checked with the microscope, using the low power lens. These dilutions usually showed definite agglutination and were considered plus-minus, i.e., if the dilution showed macroscopic agglutination in a 1:2,560 dilution and microscopic agglutination in a 1:5,120 dilution, the final agglutination was considered 1:2,560 plus.

After adding the drop of bacterial suspension to the dilution of the serum, the final dilution ranged from 1:20 to 1:10,240. The control rabbit serum showed no agglutination. The inoculated rabbit showed marked microscopic agglutination in the 1:2,560 dilution plus.

Two days later or six days after the last inoculation the rabbit was bled from the heart. The technic is as follows: The rabbit was not fed 24 hrs. previous to bleeding. A very satisfactory animal board was made of ply-wood 20 x 36 inches. About 2 inches from the corners, four brass hooks were placed. Pieces of tape 24 inches long were doubled and were placed on the rabbit's legs with a slip knot. This procedure required two workers. After the tapes were applied, the two workers turned the rabbit on its back and proceeded to tie the tapes to the hooks. It is necessary to stretch the animal out as much as possible so that it cannot twist while it is being bled.

The fur was clipped from around the heart area and was painted with tincture of iodine. Alcohol was then applied to help lay down the surrounding fur. A sterile 50 cc. syringe was assembled and a  $3\frac{1}{2}$  inch piece of rubber tubing with a 19 gauge needle attached, which had been previously sterilized by boiling for 10 mins., in 1% sodium carbonate solution, was fastened to the syringe by means of flamed forceps. To prevent clotting while the blood was being drawn, the assembled apparatus was rinsed with sterile 1.7% buffered sodium chloride solution drawing a quantity into it and then carefully expelling it. The reason for using the rubber tubing instead of attaching the needle directly to the syringe is obvious. It allows more play as the needle is being plunged into the heart. There is less chance of the needle slipping out of the heart and further damaging the animal. At this stage, one worker placed the needle into the heart and the other worker drew the plunger of the syringe rotating it and pulling firmly and evenly. The amount of blood withdrawn depended on the weight of the rabbit. A 2500 gm. rabbit can stand the loss of about 50 cc. of blood. The blood was quickly transferred to two 50 cc. capacity, round bottom centrifuge tubes with narrow mouth. Although the blood was placed in the tubes quickly, it is well to allow it to run down the side of the tube and prevent bubbling. The blood hemolyses very easily. These precautions must be observed because serum with any tinge of hemolysis cannot be used.

The rabbit was released from the board as soon as possible and was given an amount of 5% glucose in saline equal to the amount of blood drawn. It was injected intravenously. The glucose acts as a stimulant and prevents shock and mortality.

The blood was allowed to stand at room temperature for about 1 hr. to permit clotting. The clots were then carefully loosened with a stiff wire which had been flamed and cooled. After the stoppers, which were made of cotton covered with gauze, were securely fastened to the top of the tubes with elastic bands, they were centrifuged for 20 mins. The serum was carefully decanted with a sterile pipette into another sterile centrifuge tube and was again centrifuged for 10 mins. to make sure the serum was free from cells. As it was decanted this time, it was transferred to a sterile graduate and measured. As a preservative, 0.05 cc. of a 3% stock phenol solution was added per cc. of serum. The serum was trans-

ferred to sterile 30 cc. vaccine bottles with rubber caps. During this process, a small quantity of the serum was tested for sterility and another small portion was kept aside for titration. The serum, if sterile and of a high titre, was ready for use. This particular rabbit serum gave a titre of 1:5,120. Approximately 45 cc. of blood were taken from the heart with a final yield of 26 cc. of serum.

Twelve rabbits were immunized according to the above described technic. The inoculations and bleedings were carried out on the same days each week, namely Tuesday and Friday. Two rabbits were started each Tuesday and Friday until the entire twelve were receiving their doses. This arrangement worked out so that not more than two rabbits were due for bleeding on the same day. Because of the bulk of the routine laboratory work, this schedule had to be considered. The bacterial suspensions when needed, could be prepared on the intervening days.

The rabbits received no inoculations for one week after being bled. After that rest period, the inoculations were continued, starting with 6,000,000,000 bacteria per 1 cc. and increasing the doses up to 10,000,000,000 bacteria per 1 cc. or until their test bleedings showed a sufficiently high titre. They may be bled at intervals of two weeks.

The average amount of blood taken from each rabbit was about 45 cc. and the yield of serum ranged from 20 cc. to 31 cc. This series of rabbits' sera gave approximately the same titre, i.e., 1:5,120. We pooled the sera since it has been found that a patient may be more sensitive to the serum of one rabbit than another.

When the serum was ready for use, a sensitivity test was performed on the patient using the following technic: 0.1 cc. of normal rabbit serum in 5 cc. of sterile saline was prepared. The blood pressure apparatus was placed on the patient's arm and the systolic pressure was noted. The above mentioned serum was injected intravenously and the systolic pressure was watched for 10 mins. Since there was not a noticeable drop in pressure, the serum was given. In this test, if the systolic pressure drops below 20 mm. in 10 mins., serum is contraindicated. This test was made August 29, 1938, and the patient received 2 cc. of the serum.

On August 30th, 31st and September 1st she received 12 cc. twice a day. September 2nd, 3rd and 4th, she received 4 cc. respec-

tively, making a total of 86 cc. of immune rabbit serum.

The patient's blood serum previously showed no agglutination in any dilution. September 1, 1938, just three days after the first inoculation, the patient's serum showed good agglutination in the 1:20 and 1:40 dilution. September 6th, her serum showed only slight agglutination in the 1:20 dilution. Let it be remembered that the patient received no serum after September 4th. Unfortunately, the patient developed a severe serum sickness with all its characteristic manifestations and the serum had to be discontinued.

It is interesting to note that blood cultures taken on September 1st, 6th, 12th and 17th were negative after 10 days' incubation. All previous cultures, including the first day the serum was administered showed the usual streptococcus after three days' incubation with approximately 4 to 6 colonies per 1 cc. of blood. A severe nephritis had set in, blood cultures became positive again and the patient succumbed November 11, 1938.

In conclusion, may I say that our efforts seemed in vain but from the knowledge gathered that rabbits can be immunized against this particular organism with a resulting serum of high titre, the fact that the patient's serum did show some agglutination against the organism after three days' treatment with negative blood cultures resulting, seems worthy of mention.

With the serum on hand we hope to bring treatment immediately to the next case coming to our attention. In this particular case, a great deal of time was lost in getting started. We are now in much better circumstances to carry on the work. We know how little can be done for bacterial endocarditis and hope that sometime in the near future the serum can be administered with very favorable results.

Appreciation is extended to Dr. Morton and Mr. Roos for their valuable aid in producing this serum.

## SOME PRACTICAL ASPECTS ON BACTERIAL DISSOCIATION\*

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It is a somewhat startling fact that the great progress made in the diagnosis of disease by bacteriological means was due to a false conception of the nature of bacteria. This false conception was fostered by one of the early and eminent bacteriologists, Robert Koch and embodied the principles of monomorphism. Monomorphism implies that every species of bacterium, with a notable exception in the case of the organism causing diphtheria, occurs in a distinct and constant form and exhibits a fixed set of characteristic biochemical and physiologic activities. It was upon this premise that bacteria were classified and without this premise it is doubtful that the progress of medical bacteriology would have been as rapid as it has been.

Pleomorphism, as opposed to monomorphism, was advocated from the beginning of bacteriology. This concept embodies the beliefs that bacteria are extremely variable organisms that may occur in numerous shapes and sizes and may likewise vary considerably in tinctorial, biochemical and physiological activity. This view was early brought into disrepute by the careful work of Koch and his followers and the rather faulty technique of the advocates of pleomorphism. The result was that a basic truth was buried by the ineptness of its proponents.

At the present time, it is generally agreed that there are many different phases, including the mucoid, the smooth, the rough, and possibly a filterable stage, in which an organism may occur. All these phases have definite characteristics by which they may be distinguished but there also occurs many transitional phases that intervene and are not so easily classified. Fortunately, virulence has

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\* Read before the Arkansas State Society of Medical Technologists.

been fairly definitely associated with a particular phase though this association may vary with the genus. Thus, in the case of the *Pneumococci*, it is the mucoid phase that is virulent, and in the case of the *Staphylococci*, it is the smooth or S phase that is virulent. It is due to the fact that pathogenicity was confined to a single phase that the progress of medical bacteriology was not greatly hampered by the theory of monomorphism. However, the acceptance of this viewpoint has often been the cause of consternation on the part of technicians. It is possible that some of the "contaminations" that have confronted the worker were in reality dissociated forms of a pathogenic organism that remained unrecognized.

Variation in the morphology of an organism may lead to wasted effort or error on the part of the laboratory worker. As an example, let us consider the possibility of variation of the form of the pneumococcus. This organism is typically a lanceolate diplococcus but often occurs in chains resembling closely the *Streptococci* and at times may be indistinguishable from the *Streptococci*. It may also resemble closely the *Staphylococcus* arrangement though this is rarer than the *Streptococcus* arrangement. At this point the worker is confronted with the problem of choosing the next step—it would be possible to give a report of a greening *Streptococcus* rather than of a pneumococcus if the wrong line of action were undertaken. And in some instances even bile solubility or inulin fermentation may be misleading because of the power of the organisms to vary. Errors due to such causes are probably rare because the technician has learned through experience that such conditions do exist but the factors underlying these irregularities are not always appreciated nor recognized.

Likewise, in dealing with the colon-typhoid-dysentery group of organisms, the worker is often confronted with atypical fermentation reaction which leaves much doubt as to the actual identity of an organism. A colon bacillus may not ferment lactose or do so very slightly, but later may react quite typically. A pathogen may be identified as a member of the Genus *Shigella* but will react so individually that it is impossible to further separate it until much time and effort has been expended. Particularly is this true of the paradyentery organisms—in some instances, duplicate tubes may show opposite results though both were presumably inoculated with the same organism. These variations are due to the organism being in



an intermediate stage and thus not having a definite set of characteristics such as would be found in a more stabilized strain. Usually such difficulties resolve themselves when after further subculturing, the organism assumes a more stable and typical form. No better example of the variation of the biochemical activity of one species could be cited than the results of the work of Smith, Gottschall and Wallgren of Pittsburgh. These workers were able to obtain from a single species of *Lactobacillus acidophilus* seventeen different variants which could be separated on a basis of biochemical activity. Furthermore they could be identified as distinct species occurring among the thirty-five species listed in Bergey's Manual of Determinative Bacteriology. With these facts in mind, it is not surprising that one often has difficulty in identifying organisms on a basis of their action on sugars or similar substrates.

In addition to variable action on substrates, bacteria may also exhibit variation of their antigenic constitution. Usually, the mucoid phase is very specific and clear cut due to the presence of certain haptenes in the capsules, the smooth or S phase, being devoid of capsules, is not so specific and the R phase possesses an antigenic pattern that is quite nonspecific in its makeup and may overlap with the R phases of many related genera. This antigenic variation is of utmost importance in the selection of material to be used as antigen in the Widal type of test. The selection of an R phase or an intermediate R phase for use as an antigen may result in very low titers or even misleading results. Particularly must the antigen used for the opsono-cytophagic test for Brucellosis be watched in this respect. The *Brucella* organism may transform to a non-virulent type that is superficially quite similar to the virulent type, and, unless careful inspection is made the two types can not be separated on a basis of growth of the colonies. The non-virulent types of *Brucella* apparently have a less specific antigenic pattern and their use may lead to a false diagnosis because the phagocytes of a normal individual may show a marked capacity for engulfing the non-virulent forms whereas they will show little tendency to engulf the virulent forms in the absence of increased opsonins. Fresh and typically smooth cultures should always be used in performing the opsono-cytophagic index determinations.

No less important is the careful selection of strains of pathogens to be used as antigens in prophylactic measures. There can be little

doubt that many of the unfavorable results obtained by the use of bacterins are due to unfortunate selection of avirulent cultures of the intermediate or R type. These cultures of course do not have the constituents that are found in the M or S types and therefore cannot protect against them. Many believe that this fact accounts for the negative results reported by some on the use of pertussis bacterins.

More commonly, it is the low grade infections that are not clinically clear cut that are due to organisms of the intermediate types. These cases may give the technician a great deal of trouble but if the problem is approached from the standpoint of a pleomorphist the difficulty will generally resolve itself more quickly than if one overlooks the great ability of the organisms to vary in their form, staining reaction and actions on various substrates.

## ESTIMATION OF MILD ICTERUS

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Slight jaundice is difficult to detect by direct comparison of the serum with the dichromate standard in a colorimeter, when the serum is opalescent or hemolyzed to any extent. There also may be present disturbances of fat metabolism, such as diabetes and carotenemia, carotene, or carotenoid pigments in the serum.

The extraction of the serum by two volumes of acetone or absolute alcohol will secure the bile pigments in the supernatant fluid. If this is not clear it may be placed in the refrigerator for a few hours and centrifuged after precipitation is complete. Direct estimation of this extract with the potassium dichromate standard is easy after allowing for the dilution. Carotin and carotenoid pigments are in the precipitate and can be extracted from it by such lipid solvents as ether, petroleum ether, chloroform or carbon disulphide. This extraction must be prolonged and only after complete dehydration by the alcohol or acetone<sup>1</sup>.

The two methods may be combined by adding the alcohol and petroleum ether to the serum at the same time. In this method where the bilirubin pigment in the alcoholic solution is high and the lipochrome pigment in the petroleum ether solution is low, jaundice is present. The opposite indicates xanthosis. When both xanthosis and jaundice are present the serum bilirubin index and the lipochrome index are both above normal<sup>2</sup>.

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## FACTORS SUPPORTING THE FILAMENT— NONFILAMENT DIFFERENTIAL LEUKOCYTE COUNT\*

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Hematology is attracting more and more attention today and as a result of this, differential leukocyte counting is engaging the interest of well-trained medical technologists. With this in mind, I should like to review briefly the history of differential leukocyte counting and to describe in some detail the filament-non-filament method.

Since the neutrophilic leukocytes are the leukocytes which are responsible for the many methods of differential counting, let us review the normal mode of the development of these cells. By neutrophilic leukocytes we mean all of the cells which have neutrophilic granules in their cytoplasm.

The myeloblasts which develop from the megacaryocytes are the cells from which the neutrophilic leukocytes develop. These are in the red bone marrow and are rarely seen in the circulating blood. At this stage of development no granules are found in the cells. The nucleus is large, almost filling the cell. It is round or oval and has a cloudy, thready design. The myeloblasts develop into the promyelocytes which still are not present in the normal circulating blood. At this stage the nucleus has developed nucleoli. The promyelocytes develop into the myelocytes, characterized by the appearance of granulation in the cytoplasm. The nucleus is round in the youngest myelocytes and develops to oval or kidney-shaped in the older forms. The myelocytes develop into the metamyelocytes. The nucleus ceases to be round or oval becoming somewhat indented but has not taken on the form of a coiled band. The term metamyelocyte is not in common usage. It is the cell at a stage termed by Schilling "juvenile neutrophil."

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The neutrophilic metamyelocytes develop into the stab neutrophilic leukocyte. The nucleus of this form is a curved rod assuming various shapes which sometimes resemble letters of the alphabet. The nucleus may become narrower in some parts than in others.

The stab neutrophilic leukocytes develop into the polymorphonuclear neutrophilic leukocytes—the segmented neutrophilic leukocytes. In this form the nucleus is separated by fine chromatin filaments. It has from two to seven lobes. Sometimes the cell appears to have two or more nuclei but they are always connected by a filament of chromatin material. The older the neutrophils become, the more segments they will have. Obviously these cells have been in the blood stream a longer period of time.

From the above it can be seen why Dr. Beck reminds us that "the terms myeloblast, promyelocyte, myelocyte, metamyelocyte, stab cell and segmented cell do not represent distinct and separate cells" but impresses on us that "these names designate only various stages in the maturation of the granulocyte, which process is a chain of more or less continuous variation and hence each cell's step of maturity is best evaluated by itself. The divisions are more or less artificial and, as with any continuous variation, it is possible to subdivide them further into an infinite number of stages."

May I say at this point that it is therefore apparently not so important to differentiate between the various stages of the juvenile type of neutrophilic leukocytes as it is to differentiate between the total number of the immature forms in comparison to the number of mature cells. That is to say if there is a marked increase in the stab neutrophilic leukocytes, the juvenile neutrophilic leukocytes (metamyelocytes) and the neutrophilic myelocytes will be proportionately increased.

The history of the methods of differential leukocyte counting evolves itself around the individual's understanding of the development of the neutrophilic leukocyte and its interpretation by various hematologists. Lack of time prevents the mention of the names of all contributors but those who signalize the various stages are as follows:

Sixty years ago, in 1879, Ehrlich made possible the differentiation of leukocytes by his method of staining blood films. All of the cell types are included in this method. The stab neutrophilic leuko-

cyte and the segmented neutrophilic leukocyte are classed together as neutrophilic leukocytes. If his method of differential leukocyte counting were modified by placing the stag neutrophilic leukocyte in a separate class it would be as complete as any other method. For a quarter of a century his method of differential leukocyte counting remained the only one.

Since 1904 other methods of differential leukocyte counting have been offered—the difference in each being in the classification of the neutrophilic leukocyte. All other cells have maintained their original classification.

In 1904 Arneth proposed his method of differential leukocyte counting as a result of his observation of the maturation of the neutrophilic leukocytes from the myelocytes in the bone marrow. He classified the neutrophilic leukocyte into five major groups according to the number of lobes in the nuclei of the cells. The emphasis in this method of differentiation was placed on the maturity of the cells rather than on the immaturity. Arneth's contribution to the methods of differential leukocyte counting was that he demonstrated that the morphology of the nucleus of the neutrophilic leukocyte is an index to the maturity of the cell.

In 1911 Schilling proposed his method of differential leukocyte counting. For the past ten years this method has been widely used throughout this country. Schilling divides the neutrophilic leukocytes into four groups:

1. Myelocytes (the youngest cells)
2. Juvenile nuclears (metamyelocytes)
3. Stab nuclears (also called "staff" or "rod" nuclears)
4. Segmented nuclears (the most mature)

The emphasis in this method of differential leukocyte counting was placed on the immaturity of the leukocyte instead of on the maturity which had been emphasized previously. In connection with the Schilling method of differential leukocyte counting we have learned to think of the "shifts". An increase in the immature forms of the neutrophils is termed a "shift to the left" and an increase in the mature forms is termed a "shift to the right". Some hematologists are challenging the terminology "shift to the left" and "shift to the right" as being nondescript and discourage its use. These terms

have to do only with the written page. The German laboratory report blanks tabulate the immature forms to the left and the mature forms to the right; hence, the origin of the expression "shift to the left" and "shift to the right" which might be more scientifically termed "degree of immaturity" and "degree of maturity". Schilling's outstanding contribution to differential leukocyte counting was to call attention to a stage in the development of neutrophilic leukocytes between the juvenile neutrophilic leukocyte (metamyelocyte) and the two-lobed neutrophilic leukocyte (segmented). This is the stage at which the nucleus elongates before dividing into lobes. He placed this cell in a separate class and gave it a name—"stab" neutrophil.

Cooke and Pender proposed a method of differential leukocyte counting in which five groups of neutrophilic leukocytes are recognized: Class one—one lobed neutrophilic leukocytes including the myelocytes, metamyelocytes, and stab neutrophilic leukocytes; Class two—three, and four include the neutrophilic leukocytes having two, three, and four-lobed nuclei, respectively, and class five which includes those neutrophilic leukocytes having five- or more-lobed nuclei. In this method, as in Arneth's method of classification, emphasis is placed on the mature rather than on the immature cells. Cooke and Pender suggested a simple standard for determining the division of the nucleus. They observed that the nucleus of the neutrophilic leukocytes never completely divides and is joined either by broad bridges or by fine filamentous connections of nuclear material. Thus their contribution to differential leukocyte counting was to point out clearly what a lobed or segmented neutrophilic leukocyte is. They classify a nucleus as divided if its segments are connected by a chromatin thread.

Pons and Krumbhaar recognized that the methods so far discussed are all too complicated for routine work and offered, in 1924, their method of differentiation of leukocytes. They classed the neutrophilic leukocytes in four groups: Group one in which the nucleus is rounded—the myelocyte; Group two in which the nucleus is slightly indented—the metamyelocyte or juvenile neutrophilic leukocyte; Group three in which the nucleus is deeply indented—the stab neutrophilic leukocyte; and Group four which includes all segmented neutrophilic leukocytes. Pons and Krumbhaar's criterion

for the division of the neutrophilic leukocytes is quite clear. The emphasis is placed on the immature forms.

The object of the differential leukocyte count is to determine whether the myeloid tissue is active or inactive and to what extent.

An estimation of the number of immature forms is the most delicate method of studying the reaction of the bone marrow to stimulation. If the number of immature cells is a key to the activity of the myeloid tissue, then a differential leukocyte count that will give this information is competent. Farley, St. Clair and Reisinger, appreciating this, devised their filament-non-filament method of differential leukocyte counting.

Using Cooke and Ponder's criterion for determining segmentation, they divided the neutrophils into two classes—the nonfilamented group in which the nucleus is not divided to a point where the segments are connected by a fine filament and the filamented group of neutrophilic leukocytes in which the nucleus is developed to the point of segmentation with connection by a fine filament.

This method is simple and enables a quickly and accurately made differential leukocyte count and one which usually supplies the necessary information concerning the degree of immaturity of the neutrophilic leukocytes. Farley et al state "The test of a given clinical procedure is often the ease of its application divided by its usefulness."

Films for differential counting by the "filament-non-filament" method must be thin in order to demonstrate clearly the fine filament. They should be margin free, have a smooth appearance and under the microscope the red cells should be spread evenly, not touching one another. The films should appear yellow, not pink, against a white background. If a number of films are to be stained at one time, the name or room number of the patient should be written with pencil on the smear before staining. It will not wash off in the staining process. Any good blood stain is satisfactory. The slide should be studied by the "4-field meander" technic. This technic, while not followed by many technologists, is quite important because, by avoiding the unfavorable areas of the slide, it presents a fairer and more accurate determination of the distribution of the leukocytes. The larger leukocytes have a tendency to accumulate at the edge of the film while the smaller leukocytes lie at the center.



This method is an organized one in which four areas of the film are selected. Twenty-five to fifty leukocytes in each area are counted by moving into the film for a distance of four fields, across the film four fields, back to the edge of the film, again to the side for four fields, in toward the center for four fields, etc. Under no conditions are less than one hundred leukocytes counted. In some instances two hundred or more are classified but for routine counting one hundred are sufficient.

Farley et al give the following figures for normal percentage of non-filamented neutrophils:

High normal—16%

Low normal—3%

Average normal—8%

In the filament-non-filament method of differential leukocyte counting the immature forms of the neutrophilic leukocyte are easily distinguished from the mature forms because the division is clear cut. The myeloblasts and all other cells are classified as in any other method of differential leukocyte counting. Cells in which a separate lobe is noted but the filament cannot be detected are classified as filamented cells. Any neutrophilic leukocyte which is difficult to interpret should be classified as filamented although one rarely finds a cell difficult to interpret if the film is a good one.

The non-filamented neutrophils are the cells the lobes of which are connected by dense bands of chromatin material and those with a round, slightly indented or deeply indented nucleus. This group includes the myelocytes, the metamyelocytes or juvenile neutrophilic leukocytes, and the stab neutrophilic leukocytes. They are the immature neutrophilic leukocytes.

The percentage of non-filamented neutrophils closely parallels Schilling's percentage of shift, that is, if you have 26% of non-filamented neutrophils and make a Schilling differential leukocyte count on the same slide you will obtain a "shift to the left" between 24 and 28%.

While we are impressed with the filament-non-filament method of differential leukocyte counting, I should like to add that it must not be forgotten that the laboratory is working together with the clinician to obtain the greatest benefits for the patients. The medical

technologist must study, therefore, the various methods of differential leukocyte counting so that he or she can use that method which the individual clinician feels gives him the most valuable information. Well trained technologists can detect slight cytological deviations from the normal. When such deviations are noted, the pathologist is consulted for further study.

### *Conclusions*

1. An understanding of the normal mode of leukocytic development aids the medical technologist in intelligently differentiating neutrophilic leukocytes.
2. The history of differential leukocyte counting signalizes the stages of the understanding and appreciation of the development of the leukocyte.
3. A study of the filament-non-filament method of differential leukocyte counting demonstrates the simplicity of this method.

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## A NEW TECHNIC FOR DARKFIELD EXAMINATION\*

By THEODORE SIZEMORE, M.T.

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It has long been the accepted custom in the darkfield examination for *Treponema Pallida*, to prepare a vaseline ring on a slide or cover glass. A drop of normal saline was placed in this ring and the material from the ulcer or lesion transferred to the saline with a platinum loop. In this method the serum was diluted approximately 100 times, with the result that the chances of finding the organism proportionately reduced.

The following technic has been used by the writer for several years with much more success in demonstrating organisms than with the old accepted method.

The ulcer or lesion to be examined is cleansed carefully with sterile normal saline or Ringer's Solution, removing all debris and pus. The ulcer is then dried by the application of ether. A sufficient amount of serum for examination may be obtained by exerting general pressure upon the sides of the ulcer or lesion. Place the top of an ordinary blood coagulation pipette to the serum and tilt until enough of the serum is drawn into the pipette. Transfer to a clean slide using a small rubber bulb to expel the serum. Place a cover glass over the serum immediately and compress it slightly. Mount as usual in cedarwood oil.

There are several advantages to this method. Since there is no dilution of the serum the possibilities of demonstrating the organism are greatly increased. The film being thin and uniform, the operator has greater depth and breadth of focus. The ease of preparation and disposal of materials likely to infect the technician is also an important factor to be considered.

## A TECHNIQUE FOR A RAPID REPORT OF SYPHILIS IN BLOOD

By AURELIO A. LOPEZ, M.T.,

*Hospital Amador Guerrero, Colon, Republic of Panama.*

Before proceeding I must give credit to Dr. Benjamin Kline and Dorothy Lloyd whose article in March issue of the Supplement of The American Journal of Clinical Pathology inspired me to do some investigation on the Kahn Reaction which enables me now to perform a Kahn Reaction and make a report in between 15 to 20 minutes.

Dr. Benjamin Kline and Dorothy Lloyd discovered that by inactivating the blood serum for the Kline Test for syphilis in the blood for four (4) minutes at a temperature of 61 to 62° C. they had obtained the same results as by the inactivation for 30 minutes at 56° C.

The increase in temperature increases the inactivating power, but if heat is carried over 65° C. destroying the serum's amboceptor (which is a thermostable substance) the readings may be negative. If temperature is still carried higher coagulation may ensue.

After reading said article I thought of its applicability to the Kahn Test, and experimented on 550 bloods activating them at different temperatures ranging from 61 to 64° C. for four (4) minutes with best results at 63° C.

Upon checking these tests against those performed with the classic procedure (30 minutes at 56° C.) I obtained practically the same results with the exception of a few one plus reading with the classic procedure which gave me a negative reading with the new technique.

I would have liked to have made a provocative test on those patients to find out whether they were real syphilitic patients or not; but this was out of my control.

In conclusion this reaction is analogous to the classic Kahn Test for syphilis in the blood with the exception of the inactivating time which in this test is reduced to 4 minutes in comparison to 30 minutes of the classic test.

This modification of the Kahn Test enables one to perform the reaction in 15 to 20 minutes, e.g.:

While the antigen dilution is resting for 10 minutes the serum can be inactivating during that same period for 4 minutes; after the antigen is ripe and pipetted in each tube, the serum is ready to be added and the tubes shaken for three minutes; Saline solution is then added and tubes are again vigorously shaken and results recorded. All this does not require more than 15 to 20 minutes.

## **ABSTRACTS**

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**POLIOMYELITIS AND THE LYMPHATIC APPARATUS: J. M. Yoffey, C. K. Drinker. Jr. Exp. Med., vol. 70, No. 1, July, '39, p. 83.**

In experimentally infected monkeys the virus was not demonstrable in cervical lymph or in thoracic duct lymph and infection could not be produced by injecting virus into lymph nodes or by rubbing it into the free nerve endings in taste buds.

**THE ABORTIFACIENT ACTION OF THE SERUM AND URINE FROM PATIENTS WITH CANCER: K. W. Thompson, T. Hale, Jr., B. B. Whitcomb, Amer. Jr. Can., Vol. 37, No. 2, Oct. '39, p. 233.**

A principle which caused the termination of early pregnancy in rabbits was found in the serum or urine of most of the tested cancer subjects but, as 3 of 6 apparently non-cancerous patients gave the reaction and one of the known cancerous patients failed to give it, the test is apparently not specific.

**INFECTIONS WITH BACT. ENTERITIDIS IN INFANCY WITH THE TRIAD OF ENTERITIS, CHOLECYSTITIS AND MENINGITIS: K. J. Guthrie, G. L. Montgomery, Jr. Path. & Bact., Vol. 49, No. 2, Sept. '39, p. 393.**

A report of 28 cases, 16 of which were fatal. Fifteen of the fatal were newborn infants. Duration was 1-5 days. Spinal fluid was turbid, cells abundant and mostly polys. Coliform bacilli were present. Methods used to isolate Bact. enteritidis are given with the characteristics of the strains. Meningitis and enteritis were the common features of the outbreak with some also showing varying degrees of cholecystitis. Clinical evidence of blood spread was present in the 16 fatal cases but not in the 12 which recovered. The source of the outbreak was not determined but, it was thought to be milk-borne.

**THE ANITSCHKOW "MYOCYTE":** J. C. Ehrlich, B. Lapan. *Arch. of Path.*, vol. 28, No. 3, Sept., '39, p. 361.

Paraffin sections of hearts of a variety of animals and of human embryonal and age series were examined. The authors conclude that the Anitschkow "myocyte" is normal to the supporting tissue of the human heart in both embryonal and post embryonal stages. They suggest "myocardial reticulocyte" as a better name for this cell.

**CARCINOMA CELLS IN THORACIC AND IN ABDOMINAL FLUIDS:** M. J. Schlesinger. *Arch. of Path.*, vol. 28, No. 3, Sept., '39, p. 283.

Technic is given for preparation of fluids. Stained or unstained smears were found unreliable. Sections should be reported positive only when groups of polygonal cells showing polarity, sharply distinct cell walls and acinar or pseudoacinar formation are found. About 60% of fluids accumulating as the result of carcinoma were found to give these results.

**THE PROTEIN TYROSIN REACTION. A BIOCHEMICAL DIAGNOSTIC TEST FOR MALARIA:** H. O. Proske, R. B. Watson. *Jr. of Med.*, vol. 20, No. 7, Sept., '39, p. 279.

The authors discuss the Henry test with its limitations. The technique of the protein tyrosin test is given, based on the tyrosin values for serum euglobulin. Values indicative of malaria are reported in 97.4% of known malaria as compared with 81.9% positive findings by the thick blood film method. Since the test is not specific, it is not offered to supplant the stained smear but, as a diagnostic aid.

**A NEW CARBOHYDRATE FOR PREVENTION OF NUTRITIONAL ANEMIA IN INFANTS:** C. L. Wilbar. *Am. Jr. Dis. Child.*, vol. 58, No. 1, July, '39, p. 45.

By clarifying sugar cane syrup by settling and decanting and filtering through a Buchner funnel instead of with the use of carbon, it was possible to retain sufficient Fe and Cu to meet the infants' need by adding this to the formula.

**DIFFERENTIAL BLOOD PICTURE AND TOTAL CELL COUNT ON NORMAL AND TRICHINA INFECTED ALBINO RATS:**

**E. H. Beahm, C. M. Downs, Jr. Parasitology, Vol. 25, No. 5, Oct. '39, p. 405.**

The authors summarize the findings other workers have reported on a variety of animals. The method of infection is given. RBC were unaltered. WBC increased with the severity of the infection showing a relative, followed by absolute neutrophilia. As more lymphocytes entered the blood, relative neutropenia developed. The eosinophilia characteristic of trichina infection in man and hogs was not found in the rat.

**SOME PROBLEMS OF DIPHTHERIA IMMUNIZATION: M. M.**

**Hillman, P. H. & J. I. Linde, Jr. Ped., Vol. 15, No. 4, Oct. '39, p. 513.**

After a survey of Schick tests following various immunization techniques the authors report a larger percent of negatives among males than among females.

They conclude that as the number of negative Schick reactions is less 1 yr. after immunization than 6 mos. after, the test is more valuable when performed at the end of a year. Three doses of toxoid were found superior to two.

**STUDIES ON THE MENINGOCOCCUS AND MENINGOCOCCUS INFECTION: N. Silverthorne, J. G. FitzGerald and D. T. Fraser,**

**Jr. Ped., Vol. 15, No. 4, Oct. '39, p. 491.**

Fifty-one cases were studied with 42% mortality. Organisms were isolated from spinal fluid, naso-pharyngeal cultures and blood smears of purpuric lesions.

Contacts in these homes showed 31% positive naso-pharyngeal cultures as compared with 20% positive in a survey of normal healthy non-contacts.

**BLOOD GROUPS IN DISPUTED PATERNITY: M. Pijoan, Am. Jr. Med. Juris., Vol. 1, No. 1, Sept. '38, p. 5.**

The author discusses the inheritance of blood agglutinogens A, B, M & N and their Mendelian dominance. Iso-agglutination reactions have been accepted as evidence in medico-legal practice in a



number of countries.

Blood grouping has also been used in criminal identification, for example, blood found on a suspected car was found to have the same agglutinogens as that of the victim.

**THE CLINICAL VALUE OF QUANTITATIVE VITAMIN DETERMINATIONS:** T. T. Mackie, W. H. Eddy, R. Bach, *Am. Jr. Dig. Dis.*, Vol. 6, No. 9, Nov. '39, p. 617.

Vitamin assays in ulcerative colitis and a variety of nutritional diseases have convinced the authors that the methods and normal standards are sufficiently accurate for clinical work and as a guide to therapy.

**A STUDY OF THE HAZARD FROM TUBERCLE BACILLI IN ENVIRONMENTAL AIR:** R. P. Sim, F. B. Flinn, *Am. Jr. Hyg.*, Vol. 30, No. 3, Nov. '39, p. 135.

A group of experiments performed with guinea pigs and cultural methods in TB sanatoria showed that as the tubercle bacillus is rather large, it falls rapidly and that as it is coated with albuminous material when expelled from the mouth, it tends to remain where it falls and is not recirculated in the air.

**THE FORMATION AND MOVEMENTS OF LYMPH:** C. K. Drinker, *Am. Heart Jr.*, Vol. 18, No. 4, Oct. '39, p. 389.

Description of direct experiments on the lymphatic system with plates and diagrams.

**CLINICAL STUDIES IN ACIDOSIS AND ALKALOSIS; USE AND ABUSE OF ALKALI IN STATES OF BICARBONATE DEFICIENCY DUE TO RENAL ACIDOSIS AND SULFANILAMIDE ALKALOSIS:** A. F. Hartman, *Ann. Int. Med.*, Vol. 13, No. 6, Dec. '39, p. 940.

Data and cases demonstrating  $\text{BHCO}_3$  reduction in chronic nephritis, failure of the tubules to reabsorb  $\text{BHCO}_3$  and the efficacy of treatment with sodium—lactate.

The author maintains that the  $\text{BHCO}_3$  reduction following sulfanilamide has been incorrectly interpreted and that it is compensatory for a  $\text{CO}_2$  deficit alkalosis and therefore the routine administration of alkali with sulfanilamide is erroneous.

**TECHNIQUE OF SPUTUM EXAMINATION:** J. E. Pottenger, *Am. Rev. of Tub.*, Vol. 40, No. 5, Nov. '39, p. 581.

The probability of demonstrating tubercle bacilli in positive cases is discussed. Dilution-flotation technique was found adequate when present to about 175 per cc. of specimen with a 10 min. search. A guinea-pig inoculation with a 24-hr. inoculum was insufficient. A 3-day inoculum was found positive in 66.1 % with positivity increasing until a 15-day inoculum gave positive findings in 86 %.

**BACTERIOSTATIC POWER OF SERUM IN PULMONARY TUBERCULOSIS:** W. A. Kreidler, C. W. Nissler, *Am. Rev. Tub.*, Vol. 40, No. 5, Nov. '39, p. 604.

A method is described for studying the inhibitory effect of constituents of the patient's serum on the strain of bacilli isolated from the sputum. It is suggested as an aid in determining prognosis or advisability of surgical procedures.

**A SEROLOGICAL REACTION IN TUBERCULOSIS:** J. B. West, R. M. Easterling, *Am. Rev. Tub.*, Vol. 40, No. 5, Nov., '39.

A preliminary report of experiments on the factors involved in a serological diagnosis of tuberculosis. Study was based on the work of Hinton.

**EFFECT OF VISCOSITY OF SERUM ON THE RATE OF ABSORPTION OF ANTIBODIES:** D. C. Lahiri, *Ind. Jr. Med. Res.*, Vol. 27, No. 1, July '39, p. 225.

Antidiphtheric horse serum was found to be absorbed less rapidly when its viscosity had been increased by the addition of various substances. The difference in absorption between these preparations and those of saline bases diminishes until after 48 hours they are about equal.

**THE INFLUENCE OF VARYING LEVELS OF CALCIUM INTAKE ON THE BIOLOGICAL VALUE OF PROTEINS:** M. Swaminathan, *Indian Jr. Med. Res.*, Vol. 27, No. 1, July '39, p. 147.

Using the nitrogen-balance and growth methods as criteria, the values of a mixture of casein and rice proteins were not influenced by calcium concentrations ranging from 0.036 to 0.4 %.

## BOOK REVIEWS

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**A TEXTBOOK OF LABORATORY DIAGNOSIS** With Clinical Applications for Practitioners and Students. By Edwin E. Osgood, M.A., M.D., Associate Professor of Medicine and Head of the Division of Experimental Medicine, University of Oregon Medical School and Member of the Staff of Multnomah County Hospital and the Consulting Staff of the Doernbecher Memorial Hospital for Children, Portland, Oregon. Third edition with twenty-seven figures in the text and ten colored plates. 653 pages. Published January 2, 1940. The Blakiston Company, 1012 Walnut Street, Philadelphia, Pa. Price \$6.00.

The subject matter in Part One of this volume is presented in a way that closely correlates the clinical with the laboratory side of practice. This makes the text not only informative but most interesting. Instead of simply presenting cold, hard laboratory facts and technics as such, the author takes you behind the scenes and throughout the work points out the reasons and rationale for the various laboratory procedures in correlation with the clinical picture. Interest is further enhanced in the arrangement of the subject matter by systems. In Part One for example the various chapters take up the disorders of the kidney and urinary tract with especial reference to nephritis; disorders of carbohydrate, protein and fat metabolism with especial reference to diabetes mellitus and disturbances of acid-base equilibrium; disorders of the gastro-intestinal tract; disorders of the erythropoietic, the leukopoietic and the hemostatic systems, etc. Information is given which enables the reader to determine what laboratory procedures are indicated to aid in making a diagnosis, how often they should be repeated, what conclusions he is justified in making from the laboratory report and whether his treatment is producing results or doing harm. The student or clinician is thus enabled to put the proper evaluation on laboratory reports. Part One is a well correlated physiology, clinical pathology, differential diagnosis and an interpretation of the underlying pathology from laboratory findings.

Part Two is a consideration of the principles and technics of laboratory procedures. Not all technics for each test are given but those which the author uses and has found most practical are given in detail as well as others which are commonly used. Many minor modifications are included, which, through extensive use, are considered improvements. The bibliography has been planned to supplement and extend the information given, to guide the reader to the important literature with a minimum expenditure of time and for other pertinent reasons.

**MASSAGE AND REMEDIAL EXERCISES in Medical and Surgical Conditions.** By Noël M. Tidy, Member of the Chartered Society of Massage and Medical Gymnastics; T.M.M.G.; late Sister-in-Charge of the Massage Department, Princess Mary's Royal Air Force Hospital, Halton. Fourth edition, 1939. Pp. 458. A William Wood book published by The Williams & Wilkins Company, Baltimore, Md. Price \$5.25.

In the preparation of this comprehensive work the author has drawn largely from her extensive experience in the field of physiotherapy. A detailed description of physiotherapeutic methods is given for every medical and surgical condition or disease in which such methods are of value. Fractures and dislocations are dealt with in detail. In this fourth edition two short sections have been added on myositis ossificans and on the effects of gas poisoning. Other conditions are far too numerous to list here but mention of a few of them will give some idea of the scope of the work: anaemia, hemophilia, purpura; diseases of the respiratory organs as lobar and broncho-pneumonia, bronchiectasis, tuberculosis, asthma, pleurisy, empyema and many others; abdominal and pelvic conditions such as peptic ulcer, colitis, enteritis, constipation, disturbances of menstruation, enuresis, marasmus, etc.; other chapters take up heart diseases, blood and lymphvessel diseases, the various form of arthritis, deformities, functional and organic nervous diseases and many others.

This work serves as an excellent guide and text for the graduate physiotherapist. It is a valuable source of information for the physician who directs or supervises the work. This responsibility

properly belongs to the physician in charge and should not be shifted entirely to the physiotherapist. There is one criticism that should be made, namely, that a glossary explaining the terms used for the various exercises and positions should be included. This would enhance the value of the work for those unfamiliar with them and perhaps also for those who may use other terms for describing the same exercises and positions. Otherwise the book is highly recommended.

## NEWS AND ANNOUNCEMENTS

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### PRESIDENT'S MESSAGE

To the members and guests of the American Society of Medical Technologists:

We have read and heard of the plans for our eighth annual convention to be held in New York City on June 10, 11 and 12. To those of us who have attended several previous conventions, the program and the exhibits appear better than ever before. Our meeting will be completed with scientific program exhibits and entertainment. We shall come to learn, to see, and to compare. We shall see old friends, and easily make new friends. We shall be better medical technologists because of our attendance. Won't you join us in New York?

Sincerely,

BERNICE ELLIOTT,

*President, A.S.M.T.*

Additional appointments to the list of Sub-Counsellors:

District of Columbia, Miss Margaret Bush, Washington, D. C.  
North Carolina, Miss Rodwell Hunter, Raleigh, No. Car.  
Mississippi, Mrs. Sue McCarthy, Electric Mills, Miss.  
Georgia, Miss Mary Broderick, Savannah, Ga.  
Wisconsin, Miss Marian Tuttle, Superior, Wis.  
Louisiana, Miss Margaret Azcona, Alexandria, La.  
Arizona, Mr. Theodore Keiper, Tucson, Ariz.

During the convention of the American Medical Association in New York City, June 10 to 14, 1940, the Jefferson Medical College Alumni Association will hold its Reunion Banquet on Wednesday, June 12, at 7 o'clock P. M., at the Murray Hill Hotel on Park

Avenue at 40th Street. Tickets are \$2.50 each.

Request for reservations may be addressed to me at that hotel.

But if you neglect to make reservations—come anyway.

THOMAS F. DUHIGG,

*Chairman Dinner Committee.*

### NATIONAL

Massachusetts Institute of Technology, Cambridge, Mass., Department of Biology and Public Health, is again offering summer courses in General Bacteriology (June 10 to June 28, 1940) and Public Health Bacteriology (July 1 to July 19, 1940). Hours in each course, 9:30 A. M. to 12:30 P. M. and 1:00 P. M. to 4:00 P. M. For further information concerning registration for the above subjects, send all communications to Professor John W. Williams, of the Department of Biology, Public Health, Massachusetts Institute of Technology, Cambridge, Mass.

### *Oklahoma*

The Oklahoma Society of Medical Technologists held its annual spring meeting May 4, 1940, in the Mayo Hotel, Tulsa, Oklahoma.

The afternoon was devoted to a Symposium on Laboratory tests used in the Diagnosis and Treatment of Pneumonia lead by Dr. William H. Baily, Pathologist at Wesley Hospital, Oklahoma City, Oklahoma.

The following program was presented:

"The Blood Count in Pneumonia," Mary Sherry, M.T., Wesley Hospital, Oklahoma City.

"Suitable Media for Blood Cultures," Gene Sewell, M.T., University Hospital, Oklahoma City.

"Methods for Concentrating Organisms in Sputum," Marie Clark, M.T., Medical Arts Laboratory, Oklahoma City.

"The Use of Mice in the Neufeld Typing," Lucille B. Wallace, M.T., Department of Bacteriology, University of Oklahoma Medical School, Oklahoma City.

"Sputum Typing with Cultures in Infants," Zana Skidmore, M.T., St. Johns Hospital, Tulsa, Oklahoma.

"Sulphamethythyaiol in Infant Pneumonia," Vernal Johnson, University Hospital, Oklahoma City.

"Sulphapyridine Determinations," Jessie Meador, M.A., University Hospital, Oklahoma City.

"Percentage of Typing in Clinical Pneumonia at University Hospital," Ann Sandos, University Hospital, Oklahoma City.

"Incidence of Other Organisms," H. Spencer, City Health Laboratories, Tulsa, Oklahoma.

Dr. I. A. Nelson, Pathologist to St. John's Hospital, Tulsa, Oklahoma, and past concilor of the American Society of Clinical Pathologists, spoke on "Medical Technology under Current Trends" at a Banquet held at 7:30 P. M., at the Mayo Hotel.



**American  
Society of  
Medical  
Technologists**

**Program of the  
Eighth Annual Convention**

Headquarters  
*The BILTMORE*  
Madison Avenue at 43rd Street  
New York City

JUNE 10, 11, 12, 1940





### COMMITTEE CHAIRMEN

- Program*—DORIS BOWMAN GRIFFITHS, Utica, New York.  
*Exhibits*—ANNETTE CALLAN, Southern Pines, North Carolina.  
*Publicity*—DAVID SILCOCK, Versailles, Kentucky.  
*Local Arrangements*—PHYLLIS STANLEY, Newark, New Jersey.  
*Entertainment*—MARION GIANNINY, Philadelphia, Penn.  
*Sisters' Reservations and Entertainment*—SR. M. CELESTE WAGNANT, Baltimore, Md.  
*Reception and Registration*—MARIE FORTNA, Hackensack, N. J.
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**REGISTRATION**—Members and guests are requested to register upon arrival, at the registration desk, Hotel Biltmore.

**RESERVATIONS**—If you have not already made your reservations, write immediately to Biltmore Hotel.

**ACCOMMODATIONS** for the Sisters attending the National Convention have been obtained at the following locations:

- The Leo House, 330 West 23rd Street
- Sisters of Mercy, 81st Street
- St. Cecelia's Convent, 106th Street

Convention Hall, in the Biltmore Hotel, may readily be reached by Bus.

The Sisters wishing to attend the Convention, and desiring reservations, will please signify their intentions at an early date—at least three weeks prior to June 10th—by communicating with one of the members of the following committee:

- Sister M. Magdalen Bero, Mercy Hospital, Watertown, N. Y.
- Sister M. Theophane, St. Francis Hospital, Charleston, W. Va.
- Sister M. Celeste, Mercy Hospital, Baltimore, Md.

**TRANSPORTATION**—For transportation, see your local agent for fares and routes.

# American Society of Medical Technologists

## EIGHTH ANNUAL CONVENTION

HEADQUARTERS, *The BILTMORE*  
NEW YORK

JUNE 10-11-12, 1940

Registration June 10, 8:30 A. M. to 12 M.

Exhibits Open 12-2 and 4-9 P. M. Daily

MONDAY MORNING, JUNE 10, 1940, 9:30 A. M. to 12 M.

*Presiding*—DORIS B. GRIFFITHS, Utica, N. Y.

### OPENING SESSION

INVOCATION by the Reverend Thomas A. Sparks  
of the Cathedral of Saint John the Divine,  
New York City, New York.

### ANNOUNCEMENTS

PRESIDENT'S MESSAGE—Bernice Elliott, Omaha,  
Nebraska.

1. "Twelve Years of Registry: Its Contribution to Medical Technology"—Dr. Kano Ikeda, The Chas. T. Miller Hospital, St. Paul, Minn.
2. "The Medical Technologist in Industrial Medicine"—B. Anita West, E. I. DuPont De-Nemours and Co. Laboratory, Wilmington, Delaware.

### SELECTION OF CONVENTION DELEGATES

**MONDAY AFTERNOON, JUNE 10, 2-5, P. M.**

*Presiding*—ANN SNOW, North Little Rock, Ark.

1. "Incidence of Intestinal Parasites," HERMINE TATE, Charity Hospital, Lafayette, La.
2. "Hookworm: History, Identification and Modes of Infection with Laboratory Reports in 15 Cases," MARIAN BAKER, Taylor Clinic, Lufkin, Texas.
3. "Some of the Diagnostic Problems Associated with Pinworm Infestation," DR. HARDY A. KEMP, Dean University of Vermont College of Medicine, Burlington, Vt.
4. "Echinococcus Cyst of the Liver," EDWARD P. WALKER, Johns Hopkins Surgical Path. Laboratory, Baltimore, Md.
5. "Criteria for the Identification of the More Common Intestinal Parasites with Methods for the Preparation and Examination of Specimens of Stools," DR. EMMA S. MOSS, Dept. of Pathology and Bacteriology Louisiana State University School of Medicine and Charity Hospital of Louisiana at New Orleans.
6. Examination of Drawings of Intestinal Parasites loaned through the courtesy of DR. EDWIN S. GAULT, Assoc. Professor of Pathology and Bacteriology, Temple University School of Medicine, Philadelphia, Penn.

**MONDAY EVENING, JUNE 10**

(Refer to Entertainment program, page 141)

**TUESDAY MORNING, JUNE 11, 9 A. M. to 12 M.**

*Presiding*—DOROTHEA ZOLL, Philadelphia, Pa.

1. "Laboratory Technic in Medical Mycology,"  
Dr. Elizabeth Pinkerton, Ph.D., New York  
City, N. Y.
2. "Blood Grouping and Blood Transfusions  
with Special Reference to Some of the  
More Recent Developments in These  
Fields," DR. ALEXANDER S. WEINER, Ser-  
ologist and Bacteriologist to the Office of  
the Chief Medical Examiner of New  
York City, New York.
3. "Five Years of Syphilis Serology," MRS.  
MARGARET R. HARRISON, Assoc. Chemist  
U. S. Public Health Service Ven. Disease  
Research Laboratory, U. S. Marine Hos-  
pital, Staten Island, New York.
4. "The Function of the Laboratory in the Diag-  
nosis of the Blood Dyscrasias," DR. WM.  
P. MURPHY, and ISABEL HOWARD, Bos-  
ton, Mass.
5. "Tests Employed in the Study of Allergic  
Reactions," MOLLIE HILL, Veteran's Ad-  
ministration Facility, Aspinwall, Pa.
6. "Classification of Strains of Candida (Mon-  
ilia) Isolated from Sputum," HELEN M.  
KETCHUM, Michigan State Sanatorium,  
Howell, Michigan.
7. "Technique Using Oxalated Blood in Routine  
Counts," EVELYN M. JARDINE, Mary  
Hitchcock Memorial Hospital, Hanover,  
New Hampshire.

**TUESDAY, JUNE 11, 1:00 P. M.**

Refer to Entertainment program, page 141

## TUESDAY AFTERNOON, JUNE 11, 2:00 P. M.

### SESSION OF THE HOUSE OF DELEGATES

It is suggested that our members visit the Exhibits of the American Medical Association. They are very worthwhile and a pleasant and highly enlightening afternoon is insured all who attend.

*Entertainment—Members are advised to follow entertainment program, page —, if not in session with the house of delegates.*

Please return in time for our evening activities at 8:30 p. m.

## TUESDAY EVENING, JUNE 11

Refer to entertainment program, page 141

## WEDNESDAY MORNING, JUNE 12, 9 A. M. to 12 M.

*Presiding—*EDWARD P. WALKER, Baltimore, Md.

1. "Red Blood Cell Counts—Photoelectric Colorimeter — Haemacytometer," CECELIA M. KORTUEM, St. Vincent's Hospital, Chicago, Ill.
2. "Rapid Determination of Blood Specific Gravity as an Aid in Surgery," DR. CHAS. R. DREW, Dept. of Surgical Pathology, Columbia University, College of Physicians and Surgeons, New York City, New York.
3. "The Detection of Sugar in Urine," PHYLLIS STANLEY, Presbyterian Hospital, Newark, New Jersey.

4. "The Use of Literature, Possession of an Adequate Library, the Use of Journals, Filing of References, Exchange of Copies of Methods," DR. LOUISE D. LARIMORE, Pathologist, The Greenwich Hospital Association, Greenwich, Conn.
5. "Liver Functional Tests: Their Physiological Basis," DR. FRANK KONZELMANN, Temple University, School of Medicine, Philadelphia, Penn.
6. "Toxicology in the Hospital Laboratory," DR. A. V. ST. GEORGE, President-elect of the A. S. C. P., Bellevue Hospital, New York City, N. Y.
7. "The Role of the Medical Technologist in the Post-Mortem Investigation of Disease," DR. THEO. CURPHEY, Chief Medical Examiner, Nassau County, Meadowbrook Hospital, Hempstead, Long Island, N. Y.

#### WEDNESDAY AFTERNOON, JUNE 12

Refer to Entertainment Program, page 141

#### WEDNESDAY EVENING, JUNE 12

##### ANNUAL BANQUET

Refer to Entertainment Program, page 141

## THE ENTERTAINMENT PROGRAM

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### MONDAY, JUNE 10

#### 6:30 P. M.—Dinner at Bonat's.

*This will be an informal dinner at Bonat's, 331 West Thirty-first Street, a French restaurant, serving dinner in the true Provincial style.*

#### 9:00 P. M.—Informal Reception at The Biltmore.

*At nine in the Music Room of The Biltmore there will be an informal reception for members of the Board of Registry, the American Society of Clinical Pathologists and other friends. Two movies will be shown—"Flags of the Air" through the courtesy of the American Airlines and "The Laboratory" through the courtesy of the American Society of Clinical Pathologists.*

### TUESDAY, JUNE 11

#### 1:00 P. M.—Luncheon at The Biltmore.

*A "souvenir" luncheon will be held at The Biltmore. It will be followed by a short scientific program including the newest film on Pneumonia shown through the courtesy of the Lederle Laboratories. The remainder of the afternoon is left for visiting the exhibits of The American Medical Association and sight seeing.*

#### 9:00 P. M.—Radio Broadcast.\*

*Four members of our Society will meet four members of the American Society of Clinical Pathologists on the Battle of the Sexes program featuring Julia Sanderson and Frank Crumit over Station WEAJ at 9:00 p. m.*

*After the broadcast will be a fine time to visit Radio City, the International Gardens and see New York from the Observation Roof. Others of the group will visit the World's Fair.*

*\*If you wish to attend the broadcast write to The Molle Shaving Cream Program, care National Broadcasting Company, New York, N. Y., requesting tickets for the program of June 11th, for we have been able to obtain only a limited number of tickets.*



## WEDNESDAY, JUNE 12

### 1:00 P. M.—Visit to the Lederle Laboratories.

*Those registered with the A.S.C.P. and A.S.M.T. will be the guests of the Lederle Laboratories on a trip to Pearl River. This will include a motor trip through Manhattan, crossing the Hudson over the George Washington Bridge, travelling north through the rolling country of New Jersey and back to New York at Pearl River. After a most interesting trip through the laboratories, we will be served a snack to tide us over until our return to The Biltmore about six.*

### 7:00 P. M.—Reception at The Biltmore.

*A reception is being given in the Promenade of the Music Room by The A. S. Aloe Company, St. Louis, Mo.*

### 8:00 P. M.—Annual Banquet.

*Following the reception the banquet will be held in the Music Room of The Biltmore. The awards for the best papers and exhibits will be announced. The main speaker will be Col. A. Parker Hitchins of The United States Army, now detailed to the University of Pennsylvania, who is so well known for his work in Bacteriology and Public Health. This will be the gala event of the convention. Be sure to come.*

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## SPECIAL NOTICE

If you are planning to attend the Convention, and wish to be included in any or all of the above events, kindly write Marion Gianniny, Chairman of Entertainment Committee, 143 Lewis Ave., East Lansdowne, Pa., indicating those at which you will be present. It is necessary to obtain this information that proper reservations may be made in advance.

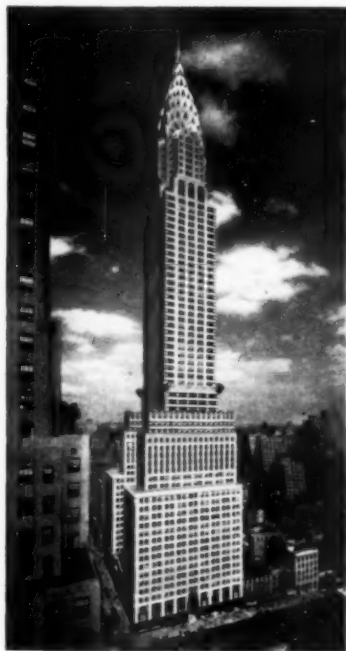
## EXHIBITS AND EXHIBITORS

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1. A—Hook Worm.  
B—A New Oxygen Tent.  
Marian A. Baker, M.T., Taylor Clinic, Lufkin, Texas.
2. Cultural Characteristics of Yeastlike Fungi.  
Helen M. Ketchum, M.T., and K. L. Burt, M.D., Michigan State Sanatorium, Howell, Mich.
3. B.J.L. Microscopic and Macroscopic Flocculation Tests.  
Marguerite Lukens, M.T., Graduate Hospital, Philadelphia, Pa.
4. Equine Periodic Ophthalmia.  
Florence Phillips, M.T., Dept. Animal Pathology, Kentucky Agricultural Experiment Station, Lexington, Kentucky.
5. Laboratory Aids.  
Phyllis Stanley, M.A., M.T., and Myrtle Craver, B.S., M.T., Presbyterian Hospital, Newark, N. J.
6. Educational Exhibit for the Laity.  
Phyllis Stanley, M.A., M.T., and S. A. Goldberg, M.D., Presbyterian Hospital, Newark, N. J.
7. A—Display on Transparent Anatomic and Pathologic Specimens.  
B—A Demonstration Exhibit on Histologic Technique for the Eye.  
Lt. Col. J. E. Ash, Medical Corps, U. S. Army and Frank M. Kramer, Army Medical Museum, Washington, D. C.
8. One Hour Renal Condition Test with Demonstration of Methods.  
W. G. Extton, M.D. and A. R. Rose, M.D., Prudential Insurance Company, Newark, N. J.
9. Manual of the Biochemical Laboratories of the Graduate Hospital of the University of Pennsylvania.  
Alexander G. Keller, Ph.G., B.Sc., Graduate Hospital, Philadelphia, Pa.
10. Fungi Influential in Disease.  
M. Elizabeth Pinkerton, Ph.D., 44 Washington Sq. South, New York City.
11. The Registry of Medical Technologists of the American Society of Clinical Pathologists, Denver, Colorado, Anna R. Scott, Registrar.

## TECHNICAL EXHIBITORS

1. Difco Laboratories, Detroit, Mich.
2. Arthur H. Thomas Company, Philadelphia, Pa.
3. A. S. Aloe & Co., St. Louis, Mo.
4. Clay-Adams Company, New York City.
5. Lederle Laboratories, New York City.
6. E. Leitz, New York City.
7. Eimer & Amend, New York City.
8. Denver Chemical Company, Denver, Colo.



THE CHRYSLER BUILDING

## NEW YORK THE CONVENTION AND VISITOR CAPITAL OF THE WORLD

### "ON TO NEW YORK"

That New York should become the convention capital of the world is wholly a natural development. The new world lavishly conferred its treasures upon the municipality. It became the metropolis of a mighty nation and, ultimately the most breath-taking city in existence. Through all this magic growth the city remained unspoiled, a neighbor to the cities of the earth, wherefore it was inevitable that New York should grow into a paradise for visitors.

The shade of the venturesome Hendrick Hudson on viewing the roof tops of Manhattan now would exclaim indignantly, "I don't believe it." Should he slip into the observation tower of Empire State Building 1,040 feet above the fairyland which is Manhattan by night, a wisp of cloud at his elbow, or should he stroll about the roof of Mr. Rockefeller's Center not far distant, doubtless he would dismiss it all as the fantastic trick of an overwrought imagination. For so it would seem and so it does seem at times to the descendants of Hendrick Hudson.

First known as New Amsterdam, New York in 1628 had 270 inhabitants. Within fifteen years eighteen languages were spoken in the town. In 1653 the municipal government was similar to that of Holland cities. Then, in 1664 the Duke of York sent out an English fleet to capture New Amsterdam and this being done bloodlessly the town was named New York after its captor.

Greater New York of today, the product of un'imited transportation facilities and of the growth of America, covers 320.03 square miles or 201,659 acres as indicated by the Typographical Bureau. As computed by the Federal Government the incorporated city covers 191,360 land acres as compared with 125,430 acres in Chicago and 81,920 acres in Philadelphia. The visitor gives most of his thought to the Borough of Manhattan which has an area of 21.9 square miles, about one-tenth the size of Chicago.

The City of New York is divided into five boroughs—Manhattan, Brooklyn, The Bronx, Queens and Richmond—whose limits are coterminous respectively with the counties of New York, Kings, Bronx, Queens and Richmond. Manhattan, the residential, commercial and financial center of the metropolis, is thirteen and one-half miles long, its maximum width being two and a quarter miles at 14th Street. The Northern and Southern extremities are only a few hundred yards wide. Prior to 1847 the city limits did not extend beyond Manhattan.

Upon the solid rock that is largely Manhattan's easy chair a city has been building whose spires pierce the clouds—when there are any clouds to pierce. Here is a city of countless wonders, a city to which the civilized world has contributed lavishly and whose attractions are so great that they draw visitors from all points of the compass.

In spite of the transformation that has taken place New York remains practically unspoiled. One of the most significant things about this Manhattan which has become the convention capital of the world is the fact that it is as friendly as a county seat anywhere—perhaps more so. Through high tide and low New York has been holding open house for the ambitious youth of the world and it is still giving them their chance. Success is still a word that anyone who wishes can learn to spell in New York. Here the so-called hardboiled are often softer underneath than those of any other city and there's a helping hand ready for any truly worthy cause.

New York attracts visitors from all parts of the world because it is not one but a thousand and one cities and each one fascinating. The city's appeal runs the gamut from the very sober to very gay. New York is a center for the arts and sciences. The richness of the past is brought together in great museums, the culture of the present is everywhere expressed.

New York day by day is a World's Fair in itself far more complete than any great exposition ever staged. Inasmuch as the Island of Manhattan is comparatively small, all of its attractions are readily accessible. Cheap transportation and plenty of it carries the visitor anywhere with a minimum of discomfort and a maximum of speed. Walking tours for shopping and sight-seeing are common because it is possible to cover the entire midtown district on foot with comparative ease.

The cost of living is roughly what the visitor wishes to make it. One can spend more or less in Manhattan than in any modern city. The visitor finds he can make a profitable visit and spend no more than he intended. The city has unexcelled hotels with accommodations priced to fit any pocket.

New York everywhere has famous and interesting buildings to show the visitor. The skyscrapers that grow thickly in Lower Manhattan and those of the midtown area stand on rock and rub noses with the sky.

Seventy-five parks and seventy-two playgrounds in the Borough of Manhattan alone add to the comfort of living; the total park system of the city amounts to 8,703 acres. Battery Park, a 21-acre area at the extreme southern tip of Manhattan, and Central Park, extending from 59th Street to 110th Street and from Fifth Avenue to Eighth Avenue, are perhaps, the two best known in New York. Bryant Park, originally a Potters Field and later known as Reservoir Square when a reservoir for the Croton aqueduct stood where stands the library today—Forty-second Street and Fifth Avenue—is a well known bit, four and three-quarter acres in size. City Hall Park, which contains city hall, the county court house, and city court is another, as are Gramercy Park, a residential park lying between Twentieth and Twenty-first Streets and owned by the abutting property owners; Madison Square, Twenty-third Street and Fifth Avenue; Bronx Park, a colloquial designation for New York Zoological Park; and, among others, the 400 acres occupied by New York Botanic Garden, adjoining Bronx Park.

Places of interest are to be found everywhere in Manhattan. In the front ranks stands the Metropolitan Museum of Art, standing in Central Park with its main entrance on Fifth Avenue and Eighty-second Street. The Museum was incorporated in 1870. It is open daily to the public. Admission is free except on Monday and Friday when a small fee is charged. In season, another institution towards which the music lover points his steps is the Metropolitan Opera, situated at Thirty-ninth Street and Broadway. A third is Carnegie Hall at 57th Street and Seventh Avenue, where the Philharmonic Symphony orchestra in season holds out with Arturo Toscanini in the conductor's stand. Summer concerts by symphony orchestras, grand opera and ballets are given under the stars at

Lewisohn Stadium in upper Manhattan. Theatres flourish the year around.

Seldom is a trip to New York thought complete without a visit to the Aquarium in Battery Park, a jaunt by steamer from the Battery to the Statute of Liberty, another to Ellis Island and Governor's Island nearby, with, perhaps, a longer voyage by sight-seeing yacht making a complete circuit of Manhattan. Jones Beach on the South Shore of Long Island is a mecca for summer visitors. This is probably the finest beach in North America.

Today's visitor to New York is much concerned with a tour of inspection of the Empire State Building, Rockefeller Center, or the Chrysler Tower. With a wisp of cloud literally at one's elbow, the visitor looks out from the observatory of the world's tallest buildings, thrilling over the panorama spread out before him. His ecstatic neighbor doubtless is just in from Australia or Buenos Aires or perhaps Canton, Vladivostok, Cape Town or Copenhagen. The endless procession of visitors to the observatories include pilgrims from every country on globe.

From these roofs, splendid views can be had of the Port of New York, admittedly the most wonderful of all harbors. Estimates have it that 75,000,000 tons of freight annually move in and out of the port by rail and 40,000,000 tons by steamship. Every twenty minutes of daylight every day an ocean going steamer comes in and one goes out. The Port of New York does not lie wholly within the limits of New York City. Army engineers taking the measurement around piers and shoreline have computed the total length at 994 miles. Steaming rapidly about the harbor, it takes two full days to cover the shore line.

New York, the home of Columbia and of New York and Fordham Universities, has a school system that recently showed an average daily attendance of 981,590 in the regular public schools and 121,500 in evening high and elementary schools. Their teachers and directors numbering 46,560 received \$134,494,162 yearly.

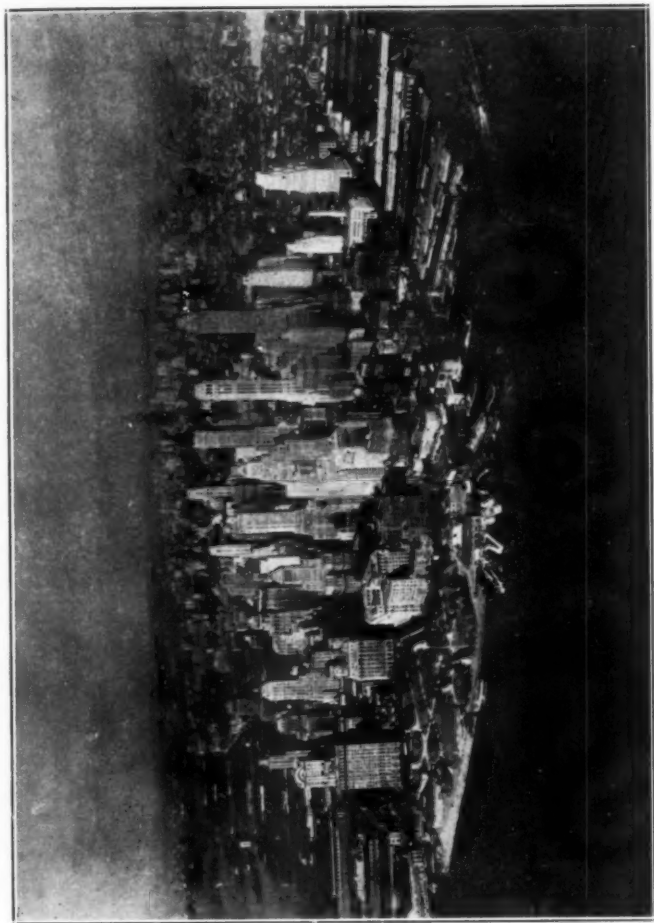
The New York stores in daytime attract the visitor as compellingly as the city's many theatres attract at night. The housewife is enchanted by the displays and it must be confessed that her husband is not far behind. After riding up Fifth Avenue on a bus, up

where the visitor can see the sparkle of the streets, the next move naturally is a tour of the stores themselves. There the people of the universe see the merchandise of the universe.

Whatever it is, as a convention visitor said, "you can do better in New York," and millions agree.

*Cuts courtesy of New York Convention and Visitors' Bureau of the Merchants Association of New York.*





An Aerial View of New York from the Harbor



THE BROOKLYN BRIDGE IN LOWER MANHATTAN

### **WARNING**

Medical Technologists are urged to caution laboratory technicians seeking certificates of qualification against joining an organization recently started in New Jersey which is not backed by any scientific or medical society but is abetted by commercial non-approved training schools.

The only institution recognized by the American Medical Association, the American College of Surgeons, the American Hospital Association, and other medical associations is the one created by the American Society of Clinical Pathologists, namely,

**THE REGISTRY  
OF MEDICAL TECHNOLOGISTS  
of the  
AMERICAN SOCIETY OF  
CLINICAL PATHOLOGISTS**

For further information address:

Mrs. Anna R. Scott, Registrar  
234 Metropolitan Building  
Denver, Colorado

